

Clinical Forms and Outcome of Tuberculosis in HIV-Infected Patients in a Tertiary Hospital in São Paulo – Brazil

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Tuberculosis (TB)/HIV co-infection significantly changes the natural history of both diseases. Proper comprehension and clinical management of co-infected TB/HIV patients is still a challenge, particularly in places like Brazil, where both types of infection are prevalent. **Objectives:** Evaluate the frequency of the clinical forms of TB in HIV-infected patients; correlate the clinical forms of TB with the level of immunodeficiency; evaluate the response to therapy with different regimens for the treatment of TB; identify potential prognostic factors in TB/HIV patients. **Material and Methods:** The following data were collected at the beginning of the study: medical history, epidemiological background, physical examination, and laboratory evaluation (complete blood cell count, T lymphocyte subsets, viral load and tuberculin test). Monthly clinical follow-up was performed, with attention to adverse reactions to tuberculostatic drugs. TCD_4^+ / CD_8^+ lymphocyte counts and quantification of the viral load were performed after 2, 4, 6, 10 and 15 months of follow-up. **Results:** The study population consisted of 78 patients (45 males and 33 females) and their mean age was 36.4 ± 7.9 years. The mean TCD_4^+ count values were higher in patients with the cavitary pulmonary form and lower in patients with disseminated forms. There were no significant differences in the mean TCD_8^+ cells counts in the different clinical forms of TB. However, the mean laboratory values for hemoglobin, hematocrit and leucocytes at study entry did differ significantly among the various clinical forms of TB. At the end of the trial, the TB recovery rate was of 78%, with four cases (5%) of treatment failure, eight (11%) treatment discontinuations and five deaths (6.4%). The highest rate of treatment failure (75%) was observed among patients with the disseminated form. Lower TCD_4^+ mean values were observed in cases of treatment failure and death. There was a correlation between the TCD_4^+ cell values and the TB outcome at the six time points. TCD_8^+ (cells/mm³) mean values assessed at the six time points in relation to the TB outcome indicated (non-significantly) lower values in patients who progressed to treatment failure. Considering the different TB outcomes, there was a significant correlation between TCD_8^+ values at the first and third assessments. Lower mean values of hemoglobin, hematocrit, platelet and leukocytes were observed among the cases of treatment failure than in patients who recovered. The variables hemoglobin, hematocrit, leukocytes and platelets were significantly different among the groups. **Conclusions:** The pulmonary forms of TB were most frequent in HIV infected patients; the extrapulmonary, associated and disseminated forms were predominantly seen in patients with a more severe level of immunosuppression. The TB recovery rate in HIV infected patients was similar to the expected rate in non-infected individuals. The best results were obtained when regimens containing rifampicin were used. Pancytopenia and low levels of TCD_4^+ and TCD_8^+ lymphocytes at the initial time point of the study were correlated with an unfavorable outcome of TB, and therefore they can be considered potential prognostic factors. However, the of TCD_8^+ lymphocyte values were the most important variable assessed.

Key Words: Tuberculosis (TB), prognostic factors, co-infection TB/HIV, HAART.

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Tuberculosis (TB) has been known for more than 6,000 years, but its control and eradication only became possible after the introduction of multi-drug chemotherapy in the 20th century. Nevertheless, the number of cases of TB has increased worldwide during the last two decades. This has occurred in part due to the human immunodeficiency virus (HIV) pandemics, since HIV coinfection significantly changed the natural balance between man and the Koch bacillus [1,2].

According to World Health Organization (WHO) estimates there are 42 million people with HIV infection worldwide, and one-quarter of these are coinfecting with *Mycobacterium tuberculosis* (MTb). This means that there are 11 million coinfecting adults and TB is the main cause of death [3-5]. Among the 23 countries with the highest incidences of TB in the world, Brazil holds the 14th position [6].

Immunocompromised individuals most commonly acquire MTb infection through the reactivation of primary latent infection (post-primary infection). The disease rarely occurs due to airborne primary infection acquired from patients with pulmonary TB [7-12].

TB/HIV co-infection significantly changes the natural history of both diseases. The hypothesis that mycobacterial co-infection increases HIV pathogenicity and speeds up progression to AIDS has been supported by an experimental model, using monkeys infected with simian immunodeficiency virus (SIV/mac) and *Mycobacterium bovis* (Calmette-Guérin bacillus – BCG) [13].

The spectrum of clinical manifestations of TB depends primarily on the level of immunodeficiency of the host. TB may occur at any stage of HIV infection [14-16] and the frequency of extrapulmonary involvement increases depending upon the severity of immunosuppression. The pulmonary form of tuberculosis is more common in the early stages of HIV infection, and the clinical and radiological findings are typical, with cavitary lesions in the upper pulmonary lobes, similarly to the findings observed in non HIV-infected patients [17,18]. Extrapulmonary forms become more frequent with increased severity of immunosuppression [15,16].

The clinical signs of the disseminated form of TB include prolonged fever, hepatosplenomegaly, anemia, pancytopenia and milliary pattern in radiographic studies. This form is also referred as the non-reactive form of the disease and occurs mainly in patients in advanced stages of immunosuppression [19-22].

Criteria for the diagnosis of TB in HIV infected patients do not differ from the criteria used for non-infected patients. However, in the context of immunodeficiency, isolation of MTb is very important due to the large number of differential diagnoses.

TB therapy regimens for HIV infected patients do not differ from the regimens used for non-HIV infected patients [6,23,24]. The combination of antiretroviral and tuberculostatic drugs can cause adverse drug interactions; the major concern is the use of rifampin, a fundamental drug for the treatment of TB. Alternative regimens without rifampin are less effective and imply the use of less active drugs, which need to administered for longer periods via a parenteral route; this may reduce treatment compliance. In these cases, antiretroviral therapy regimens based on protease inhibitors (saquinavir or ritonavir) or NNRTI (efavirenz) are preferred [25-27].

A current major concern is the increasing incidence of multidrug resistant TB (MRTB), which mainly affects coinfecting patients and must be suspected when recovery is not obtained with standard regimens [28,29].

In conclusion, proper comprehension and clinical management of co-infected TB/HIV patients is still a challenge, particularly in our environment, where both infections are prevalent. The Instituto de Infectologia Emilio Ribas (IIER), São Paulo, Brazil, is a tertiary hospital with 250 beds and an outpatient clinic, in which a large number of patients are treated. This institute is a referral center for the treatment of patients with infectious diseases, such as TB and AIDS, and it has provided relevant medical care to these patients since the beginning of the AIDS epidemics in our country. Between 1998 and 2000, 600 cases of TB were reported every year at IIER, and in 80% of these cases the HIV serologic tests were positive [30]. Considering the above-mentioned characteristics, IIER presents the necessary conditions for a study of TB/HIV coinfection.

Objectives

- To evaluate the frequency of the clinical forms of TB seen in HIV-infected patients
- To correlate the clinical forms of TB seen in HIV infected patients with the level of immunodeficiency.
- To evaluate the response to therapy using different regimens for the treatment of TB in HIV infected patients
- To identify potential prognostic factors in TB/HIV co-infected patients

Material and Methods

Patients

The study population consisted of male and female patients aged ≥ 18 years with HIV infection and recent diagnosis of TB, who were followed-up at the outpatient clinic of IIER. The screening assessments were carried out from December 2001 to December 2002. The patients were enrolled in the study only after having signed an Informed Consent.

Inclusion criteria

- Age = 18 years.
- HIV infection diagnosed by means of two positive ELISA tests and confirmed by a *Western Blot* test [26,27,31].
- Recently diagnosed TB confirmed by a positive bacteriology test (positive smear and/or culture) or histopathology.

Exclusion criteria

- Pregnancy.
- Patient lost to follow-up due to transfer or prolonged hospitalization in another service that did not allow clinical and laboratory assessment for at least three time points of the study.
- Patient refusal to sign the informed consent
- Diagnosis of non-tuberculosis mycobacteriosis.

Description of the patients

The study population consisted of 78 patients (45 male and 33 female). The mean age was 36.4 ± 7.9 years and most patients were white ($n = 62$). The average interval between the diagnosis of HIV infection and the diagnosis of TB was 4.2 ± 3.6 years. Based on the patient's answers to a standard epidemiological questionnaire regarding HIV exposure, 35 (45%) declared themselves to be heterosexual, 16 (20.5%) homosexual and 13 (17%) intravenous drug users. The risk factors could not be identified in 14 (18%) patients.

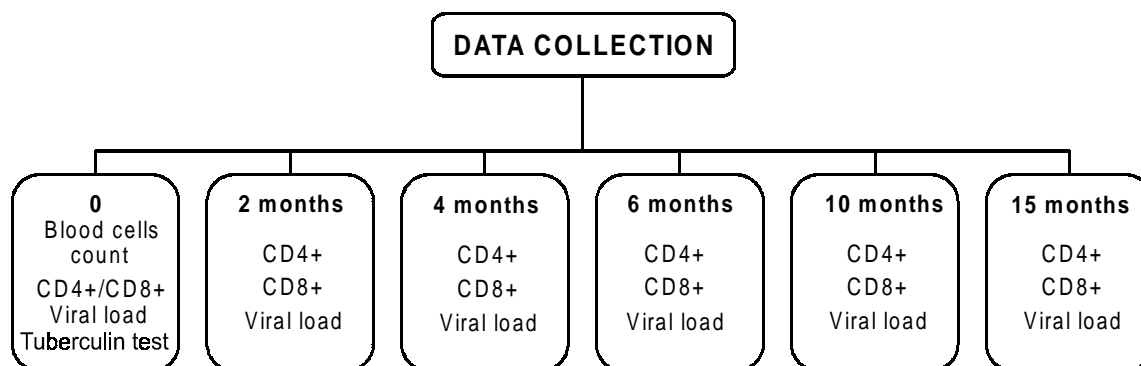
Method

The following data were collected at study entry: medical history, epidemiological background, physical examination, laboratory evaluation, including complete blood cell count, T lymphocyte subsets and viral load (measured by an HIV RNA PCR test), and a Tuberculin test. Monthly clinical follow-up was performed by a multiprofessional team, with special attention to adverse reactions to tuberculostatic drugs. TCD_4^+ and TCD_8^+ lymphocyte counts and quantification of the viral load were performed after 2, 4, 6, 10 and 15 months of follow-up (Figure 1).

Sputum smears were analyzed using the Ziehl-Neelsen (ZN) stain and were considered positive when at least 5,000 or 10,000 bacilli were detected [6,24,32]. Cultures were performed by means of radiometric tests (BACTEC 460®) and also using Lowenstein-Jensen media. Myco/Flytic media was used on the BACTEC 9000® or 9240® systems for blood and bone marrow.

Chest X-rays with an anteroposterior view were obtained and temporally related sputum samples were collected for bacilloscopic study or culture [33,34]. Imaging methods were also used for diagnosis of the clinical form of TB [33,34].

TB was diagnosed based on histological examination, by the finding of epithelioid macrophages forming confluent granulomas, giant cells of Langhans, central areas of caseous necrosis, and surrounding

Figure 1. Study assessment flow chart.

lymphocyte infiltrate. The ZN stain method was used to identify acid-fast bacilli (AFB).

A diagnosis of TB was confirmed by one or more of the following criteria: detection of acid fast bacilli using the Ziehl-Neelsen stain in sputum or in any other material; positive culture for *M. tuberculosis* in sputum or in any other material and histopathological changes consistent with TB. According to the sites where the samples were obtained, the TB was classified as pulmonary, extrapulmonary, combined or disseminated.

The pulmonary forms were classified as cavitary and non-cavitary, based on the chest X-Ray. Extrapulmonary forms were defined when TB was diagnosed in a single site, which could be pleural, urinary, lymphadenopathy, laryngeal or meningoencephalitis. Combined forms were defined when there was pulmonary involvement associated with involvement of another site. The disseminated form of TB was diagnosed when more than two sites were involved or when *Mycobacterium tuberculosis* was isolated from blood cultures in individuals with a consistent clinical picture.

Follow-up allowed classification of the cases into the following four categories, according to the outcome: recovery, treatment failure, treatment discontinuation and death.

Statistical analysis

Spearman correlations were used to determine the degree of association of the clinical forms of TB with TCD_4^+/TCD_8^+ cell counts and viral load (log10) at the six time points of the study and also the degree of association of TCD_4^+/TCD_8^+ cell counts and the viral load with the clinical outcome of TB (recovery, treatment failure, treatment discontinuation or death [35,36].

A variance analysis (ANOVA) was used to compare the means of blood cell counts at the initial assessment in relation to the clinical forms of TB and also to the clinical outcome (recovery, treatment failure, treatment discontinuation or death) as well as to compare the mean values for TCD_4^+/TCD_8^+ with the clinical outcome of TB at the six time points of the study and the mean initial TCD_4^+ values with the tuberculin test [35,36].

The ROC curve (*receiver operating characteristic*) was used to assess the possibility of identifying cut off values for the favorable or unfavorable outcomes, taking into account the following variables: TCD_4^+/TCD_8^+ cells count, hemoglobin, hematocrit and leucocytes. Logistic regression analysis was used to

assess the possibility to create a model, in which the dependent variable (TB outcome: favorable or unfavorable) was affected by a series of variables selected through Spearman correlation analysis, with a significance level of 20% (0.200 criterion for selection of variables for the regression model) [35,36]. A significance level of 5% (0.05) was used for the statistical analysis, using the software "Statistical Package for Social Sciences" version 10.0 (SPSS-10.0).

Results

Diagnosis of TB

TB was diagnosed by means of an AFB smear in 15 patients (19%). Culture alone was the diagnosis method used in 15 patients (19%); AFB smear and culture was used in 29 patients (37%); histology was used for the diagnosis in four patients (5.1%); culture and histology were used in four patients (5.1%); and bacilloscopy, culture and histology tests were used for the diagnosis in six patients (7.7%).

Clinical forms of tuberculosis

Among a total of 78 patients enrolled in the study, 28 (36%) presented extrapulmonary TB, 23 (29.5%) non-cavitary pulmonary TB, 6 (7.7%) cavitary pulmonary TB, 17 (22%) disseminated TB and 4 (5.1%) presented with both pulmonary and extrapulmonary TB.

Immunological assessment

TCD₄⁺ cells count

The mean TCD₄⁺ count values were higher in patients with the cavitary pulmonary form and lower in patients with disseminated forms, showing a different pattern during the six evaluations (Table 1).

The Spearman correlation analysis (Table 2) showed that at the six evaluations, the TCD₄⁺ cell count values varied with the clinical forms of TB.

TCD₈⁺ cell counts

The analysis of the mean values of TCD₈⁺ cell counts in the different clinical forms of TB did not reveal significant differences, and only random increases and decreases were observed. The Spearman correlation analysis performed at the beginning of the study for the clinical forms of TB and viral load values (log10) did not reveal any significant correlations.

Quantification of HIV viral load (log10)

The Spearman correlation analysis performed for the clinical forms of TB and HIV viral load values (log10) did not reveal any significant correlations between the variables.

Blood cell counts

There were significant differences in the mean laboratory values for hemoglobin, hematocrit and leucocytes (white blood cells) in the different clinical forms of TB at study entry (Table 3).

TB treatment

Regimen I (2RHZ/4RH) was initially prescribed to most patients (79.5%). Regimen I reinforced (2RHZE/4RHE) was prescribed for seven patients (9%). Five patients (6.4%) received regimen II (2RHZ/7RH) and four (5.1%) received regimen III.

The regimen was changed for seven patients who initially received regimen I. In three cases the regimen was switched to streptomycin, ethambutol and ofloxacin, due to liver toxicity. Three patients received streptomycin, ethambutol, isoniazid and pyrazinamide due to resistance to rifampin. One patient had the regimen switched to streptomycin, ethambutol and ofloxacin, due to resistance to rifampin and isoniazid.

Antiretroviral therapy

Three patients did not receive any antiretroviral therapy. In most cases (89.7%) a regimen with two

Table 1. The mean values for T CD4+ cell counts (cells/mm³) according to the clinical form at the six evaluations

Clinical forms of tuberculosis	TCD ₄ ⁺ mean values (cells/mm ³)					
	Evaluation 1	Evaluation 2	Evaluation 3	Evaluation 4	Evaluation 5	Evaluation 6
	(N = 78)	(N = 73)	(N = 78)	(N = 68)	(N = 52)	(N = 53)
Cavitary pulmonary	318.2	327.2	367.2	488.4	445.5	457.3
Non-cavitary pulmonary	209.7	260.8	291.6	286.9	338.8	402.2
Extrapulmonary	183.6	258.5	305.7	303.7	389.0	312.2
Combined**	166.7	243.2	226.5	214.7	264.5	244.7
Disseminated***	95.5	171.3	177.6	210.2	218.2	176.0
Mean	181.6	244.9	274.3	287.6	338.8	325.0

N = frequency. P = Pulmonary. ** Both pulmonary and extrapulmonary forms. *** Disseminated (hematogenic), more than two sites.

Table 2. Correlation between the clinical forms of TB and the respective values of TCD₄⁺ cells count (cells/mm³) at the six evaluations of the study

Clinical forms/ TCD ₄ ⁺ (cells/mm ³)	Correlation coefficient	P
Evaluation 1 (n = 78)	+ 0.401	< 0.001
Evaluation 2 (n = 73)	+ 0.263	0.024
Evaluation 3 (n = 78)	+ 0.259	0.022
Evaluation 4 (n = 68)	+ 0.308	0.011
Evaluation 5 (n = 62)	+ 0.270	0.034
Evaluation 6 (n = 53)	+ 0.522	< 0.001

P = significance level. A significant correlation was observed at the six timepoints.

Table 3. Mean values (\pm standard deviation) of the hemoglobin, hematocrit, platelet and leucocyte counts at study entry, in the different clinical forms of tuberculosis (TB)

Clinical forms of TB	Mean values \pm standard deviation			
	Hemoglobin	Hematocrit	Platelets	Leukocytes
Cavitary pulmonary	13.4 \pm 2.3	39.2 \pm 6.6	194,000 \pm 62,199	5,900 \pm 2,078
Non-cavitary pulmonary	11.7 \pm 2.5	34.5 \pm 7.1	226,652 \pm 103,484	4,260 \pm 1,068
Extrapulmonary	12.0 \pm 2.4	35.9 \pm 7.2	219,214 \pm 78,299	5,700 \pm 2,957
Combined*	13.0 \pm 2.7	37.0 \pm 10.3	266,250 \pm 82,163	5,525 \pm 2,692
Disseminated**	9.7 \pm 2.7	29.2 \pm 8.9	176,235 \pm 114,557	3,852 \pm 1,779
Mean	11.6 \pm 2.8	34.3 \pm 8.1	210,461 \pm 94,071	4,879 \pm 2,303
P	0.014	0.027	0.491	0.035

P = significance.* Both pulmonary and extrapulmonary forms.** Hematogenic disseminated, more than two sites.

NRTI and one NNRTI was used. A regimen combining two NTRI and two IP was used for 6.4% of the patients.

Outcome

At the end of the trial, the Tb recovery rate was 78%. There were four cases (5.1%) of treatment failure, eight (11%) treatment discontinuations and five deaths (6.4%) during follow-up.

Evaluation of prognostic factors

The highest rate of treatment failure (75%) was observed among patients with the disseminated form of TB (Table 4).

Lower TCD₄⁺ mean values were observed in cases of treatment failure and death (Table 5).

There was a significant correlation between the TCD₄⁺ cell values and the TB outcome at the six time points of the study (Table 6).

The TCD₈⁺ (cells/mm³) mean values assessed at the six time points of the study in relation to the TB outcome were lower, but not significantly, in patients who progressed to treatment failure.

Considering the different TB outcomes there was a significant correlation between T CD8+ values and TB outcome at the first and third assessments (Table 7).

A correlation between the response to the treatment and HIV viral load (log₁₀) values was observed at the end of the treatment. Among the patients with favorable response (recovery), 67% presented viral load levels < 400 copies/mm³ (log₁₀ < 2.6), and 33% presented viral load levels > 400 copies/mm³.

Among the patients with unfavorable response (treatment failure and discontinuation), 75% presented viral load levels < 400 copies/mm³ (log₁₀ < 2.6), and 25%, presented viral load levels > 400 copies/mm³ (log₁₀ < 2.6, Table 8).

In the study of a possible relationship between the TB outcome and laboratory parameters, lower mean values of hemoglobin, hematocrit, platelet and leukocytes were observed among the cases of treatment failure than in patients who recovered. In the variance

analysis performed between the means of the groups, the variables hemoglobin, hematocrit, leukocytes and platelets varied significantly (Table 9).

All cases of TB treatment failure were associated with treatments initially prescribed other than I, IR and II.

In the study of a possible relationship between the tuberculin test and mean values of TCD₄⁺ cells measured at the initial assessment, a lower number of TCD₄⁺ cells was observed in patients with a tuberculin test < 5mm. ANOVA analysis between the mean values TCD₄⁺ and the tuberculin test gave significant results.

When the ROC curve (receiver operating characteristic) was used to assess the cut off values for lymphocytes, TCD₄⁺, TCD₈⁺, hemoglobin, hematocrit and leukocytes for the different TB outcomes at the initial analysis, it was observed that the patients with < 119.5 T CD₄⁺ cells/mm³ (Figure 2), < 653.5 TCD₈⁺ cells/mm³ (Figure 3), < 10.3 g hemoglobin/dL (Figure 4), < 27.8% hematocrit (Figure 5) and < 3.2 thousand leukocytes/μL (Figure 6) were in the unfavorable outcome group.

Considering the variables selected by Spearman correlation, applied to logistic regression, the only variable capable of explaining the outcome (favorable or unfavorable) after treatment for TB in HIV infected patients was TCD₈⁺ cell count at the initial assessment.

Discussion

TB was diagnosed by the recovery of *M. tuberculosis*, Ziehl-Neelsen stain and/or culture in 74 patients. The diagnosis was confirmed by histopathological findings consistent with TB in four cases (Table 1). The basis for the confirmation of the diagnosis has remained the same since 1882, i.e. by means of isolation of the agent in culture and species identification. Cultures were considered satisfactory (69% of the cases), comparable to other referral centers [37,38].

The overestimation of bacteriological findings, based on WHO politics for the identification of new cases of TB through positive sputum smears among symptomatic

Table 4. Description of the frequency relative to the TB outcomes in 78 HIV infected patients, according to the clinical form of the disease

Clinical forms	Outcome				
	Recovery N (%)	Treatment failure N (%)	Discontinuation N (%)	Death N (%)	Total N (%)
Cavitary pulmonary	6 (9.8)	0	0	0	6 (7.7)
Non-cavitary pulmonary	20 (32.8)	0	3 (37.5)	0	23 (29.5)
Extrapulmonary	23 (37.7)	1 (25.0)	2 (25.0)	2 (40.0)	28 (35.9)
Combined*	3 (4.9)	0	1 (12.5)	0	4 (5.1)
Disseminated**	9 (14.8)	3 (75.0)	2 (25.0)	3 (60.0)	17
8888888888888888 (21.8)					
Total	61	4	8	5	78

n = number of patients. * Both pulmonary and extrapulmonary forms. ** Hematogenic disseminated, more than two sites.

Table 5. Mean TCD₄⁺ cell counts (cells/mm³) at the six evaluations in relation to the tuberculosis outcome

Outcome	T CD4+ mean values (cells/mm ³)					
	Evaluation 1 (n = 78)	Evaluation 2(n = 73)	Evaluation 3 (n = 78)	Evaluation 4 (n = 68)	Evaluation 5 (n = 62)	Evaluation 6 (n = 53)
Recovery	205.0	273.7	314.3	314.2	374.2	350.3
Treatment failure	27.0	115.5	121.5	123.5	128.5	114.2
Discontinuation	168.5	225.7	194.9	183.6	201.6	152.5
Death	40.0	46.2	35.2	0	0	0
Mean	181.6	244.9	274.3	287.6	338.8	325.0
P	0.038	0.013	0.001	0.032	0.028	0.023

N = number of patients. P = significance.

Table 6. Correlation between TCD₄⁺ mean values (cells/mm³) and tuberculosis outcome at the six evaluations

Outcome/ TCD ₄ ⁺ (cells/mm ³)	Correlation coefficient	P
Evaluation 1 (n = 78)	- 0.317	0.005
Evaluation 2 (n = 73)	- 0.358	0.002
Evaluation 3 (n = 78)	- 0.455	< 0.001
Evaluation 4 (n = 68)	- 0.367	0.002
Evaluation 5 (n = 62)	- 0.406	0.001
Evaluation 6 (n = 53)	- 0.431	0.001

P = significance.

Table 7. Spearman correlation between TCD₈⁺ cell count values (cells/mm³) and tuberculosis outcomes at the six evaluations

Outcome/T CD8+ (cells/mm ³)	Correlation coefficient	P
Evaluation 1 (n = 78)	- 0.303	0.007
Evaluation 2 (n = 71)	- 0.124	0.301
Evaluation 3 (n = 78)	- 0.232	0.041
Evaluation 4 (n = 68)	- 0.138	0.263
Evaluation 5 (n = 61)	- 0.222	0.085
Evaluation 6 (n = 63)	- 0.187	0.180

P = significance.

Table 8. Correlation between favorable response to the treatment for tuberculosis (recovery) and unfavorable response (treatment failure and discontinuation) and HIV viral load (log₁₀) at the end of the treatment

Response	Favorable	Unfavorable	Total
	N %	N %	N %
Viral load < 400* (log ₁₀ < 2.6)*	39 (67.9)	03 (25.0)	42 (60.0)
Viral load > 400 (log ₁₀ > 2.6)	19 (32.8)	09 (75.0)	28 (40.0)
Total	58 (100.0)	12 (100.0)	70 (100.0)

P = 0.009, N = number of patients. *Non-detectable viral load.

Table 9. Mean values ± standard deviation of hemoglobin, hematocrit, platelets and leucocytes values at the initial assessment, according to the clinical outcome after the treatment for tuberculosis

Outcome	Mean values ± standard deviation			
	Hemoglobin	Hematocrit	Platelets	Leukocytes
Recovery	11.9 ± 2.7	35.3 ± 8.1	222,569 ± 96,058	5,144 ± 2,370
Treatment failure	8.3 ± 1.3	24.7 ± 3.8	117,000 ± 35,721	3,075 ± 665
Discontinuation	11.7 ± 2.6	35.0 ± 7.4	225,000 ± 56,694	5,362 ± 1,575
Death	9.5 ± 1.4	28.5 ± 4.2	114,000 ± 48,270	2,320 ± 465
Mean	11.6 ± 2.8	34.3 ± 8.1	210,461 ± 94,071	4,879 ± 2,303
P	0.017	0.023	0.013	0.018

P = significance.

Figure 2. ROC curve for the T CD₄⁺ values assessed at the initial analysis according to the outcome (favorable or unfavorable) (119.5 cells/mm³).

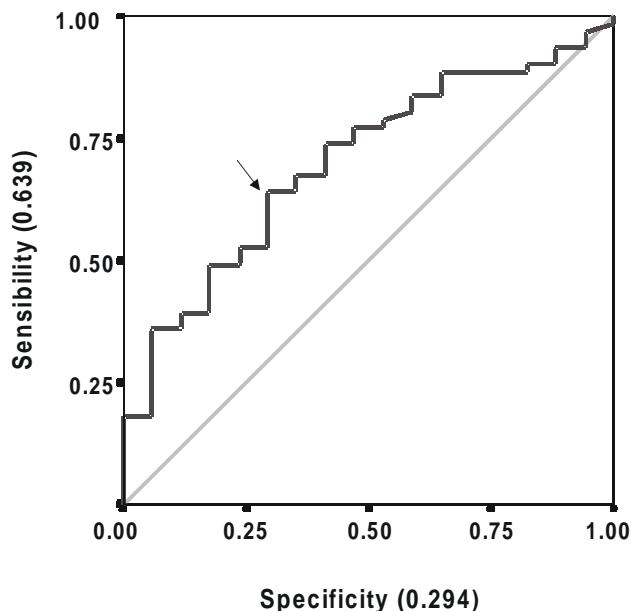


Figure 3. ROC curve for the T CD₈⁺ values assessed at the initial analysis according to the outcome (favorable or unfavorable) (653.5 cells/mm³).

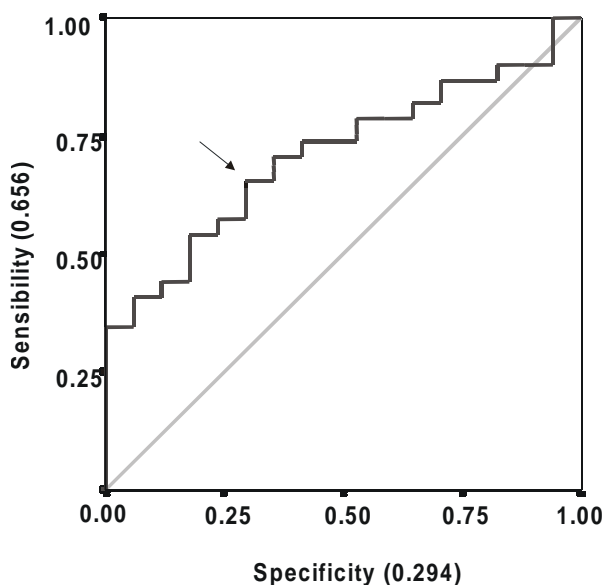


Figure 4. ROC curve for hemoglobin values assessed at the initial analysis according to the outcome (favorable or unfavorable) (10.3g/dL).

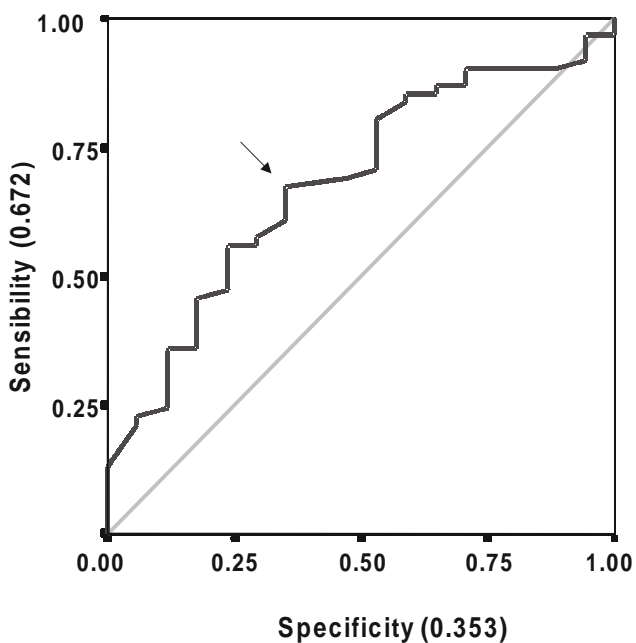


Figure 5. ROC curve for the hematocrit value assessed at the initial analysis according to the outcome (favorable or unfavorable) (27.8%).

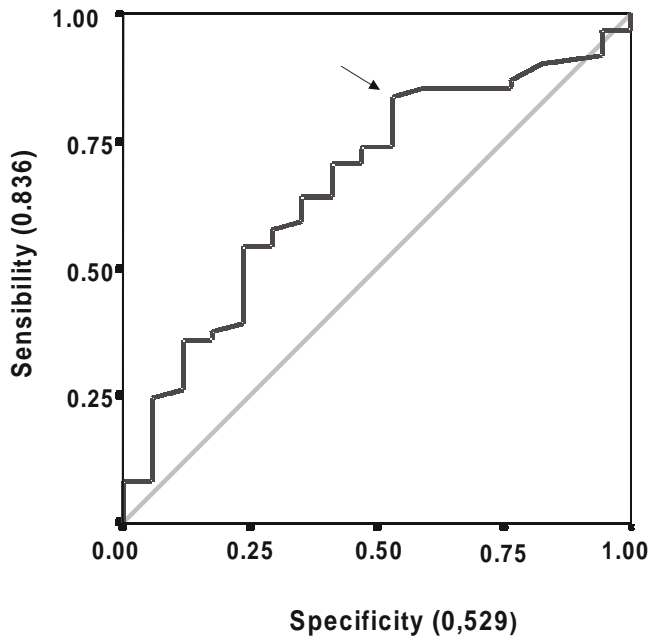
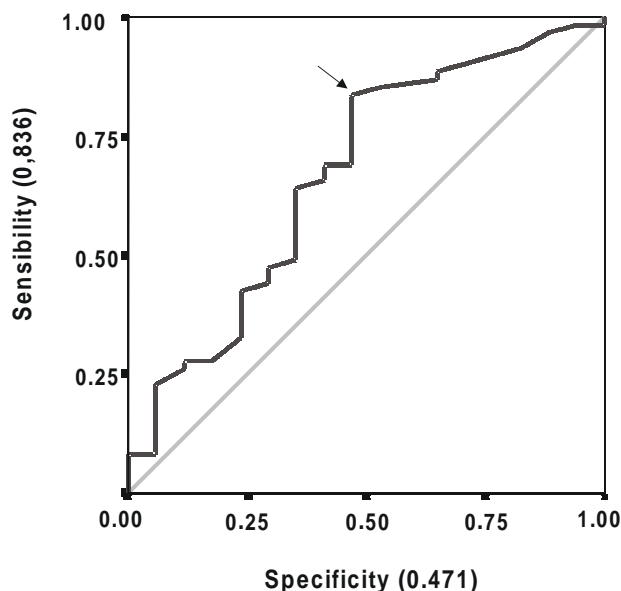


Figure 6. ROC curve for the leukocyte values assessed at the initial analysis according to the outcome (favorable or unfavorable) (3.2 thousand/ μL).



patients has been a controversial theme among physicians. Based on the new reality brought on by AIDS and due to multiresistance, there is a need to pay greater attention to the importance of radiographic findings in the diagnosis of TB [39]). In this study population, pulmonary infiltration was the main finding in the diagnosis of TB. Cavitation was seen in only six patients. These findings are similar to the ones described by Atomiya et al. (2002) in co-infected individuals.

Therefore, the two methods (isolation of the agent and radiology) are important, and they complement each other. In advanced HIV infection, which involves a large number of differential diagnoses, a thorough search for the etiological agent is needed.

The most common clinical form of TB was the isolated pulmonary form, occurring in 37% of the cases. However, the extra-pulmonary (36%) and disseminated (22%) forms occurred at a higher frequency than has been observed among HIV seronegative patients (Table 2)

Before AIDS pandemics, approximately 85% of the cases of TB were restricted to the lungs and the remaining 15% were extrapulmonary [41]. However, this distribution has changed among co-infected patients

[42,43]. In a study conducted at Complexo Hospitalar do Mandaqui (São Paulo), Melo et al. (1990) observed extrapulmonary involvement in 37.5% of the cases. The disseminated form is known to affect patients at an advanced stage of immunosuppression, and it is referred to as a non-reactive form of the disease [19-22].

We observed a significant correlation between the clinical forms of TB and mean TCD_4^+ cells count values (cells/ mm^3) in all assessments (Table 3). Among patients with the pulmonary cavitary form of TB, the level of immunosuppression was mild; a more severe level of immunosuppression was seen among patients with extrapulmonary, combined and disseminated forms of TB.

Depletion of TCD_4^+ cells and TCD_4^+ dysfunction are markers of HIV infection and are the main cause of immunodeficiency [44,45]. It is also well known that HIV infection induces alterations in the T lymphocyte subpopulations, inhibiting the protective Th1 response (IFN- γ and IL2) and inducing a non-protective th2 response (IL4, IL5 and IL10), which is involved in the production of IgE, eosinophil recruitment and inhibition of NK and th1 lymphocytes [46,47].

In contrast, *M. tuberculosis* infection may affect HIV replication and disease progression to AIDS. Several studies have reported increased rates of viral replication and decreases in TCD_4^+ cell counts during acute phases of TB [13,48-51].

TCD_8^+ cell counts had lower values in patients with disseminated TB at study entry; however, the differences were not significant. This trend was not sustained at the following assessments, as apparently random increases and decreases were observed. It is known that TCD_8^+ cell counts are reduced in patients with TB without HIV infection and are even lower in co-infected patients [52]. It was observed in a trial conducted in Brazil with 61 patients, 20 with TB, 10 infected with HIV, 14 co-infected and 17 healthy individuals, that isolated TB was the disease associated with the lowest counts of TCD_8^+ cells [53].

Discrepancies in the TCD_8^+ cell counts occur when there is interaction between these two infections (TB and HIV), which leads to TCD_8^+ apoptosis.

An increase in the HIV viral load during the acute phases of TB has been reported in several studies,

including one published by Goletti et al. (1996), who found a 5 to 160-fold elevation of the viral load during acute TB. However, we did not find significant differences among the mean values of HIV viral load (log10) in the different clinical forms of TB at the initial assessment; nor were differences evident when comparing the different clinical forms of TB and the HIV viral load values (log10) at the six time points of our study.

The more evident anemia and leukopenia seen in patients with disseminated TB may indicate a severe immunosuppression of advanced AIDS, which can facilitate hematogenous dissemination of *M. tuberculosis* [54].

Initially, 79.5% of the patients received EI (R + H + Z) and 20.5% used other regimens (EIR, EII, EIII) to treat TB. The current therapeutic regimens used in the treatment of TB among co-infected TB/HIV patients are similar to those used in non-HIV infected patients, and preference is given to short-course treatments (six months). The efficacy of these regimens among co-infected patients is usually the same due to the immune recovery obtained after the administration of a potent HAART regimen [27,55-57].

Concerning the evolution of TB, clinical recovery was observed in 78% of the patients, treatment failure in 5.1%, treatment discontinuation in 10% and death in 6.4%. In Brazil, the recovery rates were only 63.1% in 1999 [58].

Another relevant aspect of this study is that all cases of recovery were observed in patients treated with regimens containing rifampicin (I, IR and II), confirming the data available on the literature emphasizing the importance of this drug and the success rates of 90 to 95% seen with this medication [56,59]. Treatment failure was more frequently seen in the disseminated form of TB (75%); this is thought to be due to the severe immunosuppression observed in these patients.

The evaluation of the mean values of TCD₄⁺ cells in association with the TB outcome showed lower values in patients who experienced treatment failure and in cases of death. These associations were significant at the six time points of the study. The Spearman correlation test also indicated significance (Table 5).

These data emphasize the importance of TCD₄⁺ lymphocytes in the recovery from TB [46,47].

When correlating the Spearman test among the TCD₈⁺ cell counts and TB outcome (favorable or unfavorable), a significant correlation was observed at the initial assessment and at month 4 of the study (Table 7). The importance of TCD₈⁺ counts as a prognostic factor is a controversial issue, as is the increase in TCD₈ and CD₃₈ expression among co-infected patients [60]. We observed only a correlation of TCD₈⁺ count with an unfavorable outcome and not with the clinical forms of TB. This was also found by Brinchmann et al. (1990; 1994) who found that one of the functions of the TCD₈⁺ lymphocytes is to control the viremia from the beginning of HIV infection and that the number of circulating cells can be associated with the prognosis of the infection.

The correlation between the HIV viral load quantification (log10), performed at the end of the treatment, and the TB outcome (favorable and unfavorable) showed that the viral load of 75% of the patients with an unfavorable outcome was >400 copies/mL of blood (log10 > 2.6); only 32.8% of the patients who recovered presented the same results (Table 8). It is known that *M. tuberculosis* activates HIV expression [49], and that it may increase pathogenicity and speed up disease progression to AIDS [13].

The pancytopenia observed in patients with an unfavorable outcome may be considered an indication of a poor prognosis. Jamal (1998) observed in a study conducted in Brazil that in co-infected patients dissociated cytopenia or pancytopenia were significant predictive factors of early death. He also observed an association between leukopenia and death within two months after the diagnosis of TB.

In the evaluation of the tuberculin test relative to the mean values of TCD₄⁺ cells performed at the beginning of our study, it was observed that nonreactive patients had lower numbers of TCD₄⁺ lymphocytes. These results suggested that it is difficult to establish the usefulness of the tuberculin test results in co-infected patients with severe immunosuppression [64].

When the ROC curve was used to assess the cut off values for TCD₄⁺ lymphocytes, TCD₈⁺ lymphocytes, hemoglobin, hematocrit and leukocytes at the initial time

point of the study according to the TB outcome (favorable or unfavorable), the following values were obtained: TCD₄⁺ lymphocytes (119.5 cells/mm³), TCD₈⁺ lymphocytes (653.5 cells/mm³), hemoglobin (10.3g/dL), hematocrit (27.8%) and leukocytes (3.2 thousand/mL) (Figures 2-6); at lower values the outcome tended to be unfavorable, so this was considered as a prognostic factor in the co-infection (HIV/TB). Several studies have documented the importance of TCD₄⁺ as a prognostic factor. Dheda et al. (2004) showed that the risk of death is high among co-infected patients with TCD₄⁺ cells values lower than 100 cells/mm³. It is well known that the TCD₈⁺ lymphocytes are very important in the control of the viremia from the beginning of HIV infection and that the number of circulating cells can be associated with the prognosis of the infection. Jamal (1998) associated pancytopenia with early death.

However, based on the logistic regression model, the TCD₈⁺ lymphocyte count performed at the initial time point was the most important factor associated with favorable or unfavorable outcome after treatment for TB.

The study of the clinical forms of TB and TB outcome in HIV infected patients, has indicated improvement after the HAART era. However, it remains necessary to isolate the mycobacterium and prioritize conventional TB treatments. Special attention must be paid to the determination of the prognostic factors in co-infected patients, not only due to the complexity of the immunological changes seen in the TB/HIV combination, but also due to the spectrum of the immunodeficiency. Therefore, more detailed studies must be conducted to explain this.

Conclusions

– The pulmonary forms of TB were most frequent in HIV infected patients, but the frequencies of extrapulmonary and disseminated forms of TB were higher than those reported in the literature for non-HIV-infected patients.

- The pulmonary cavitary form of TB was seen in patients with mild immunosuppression, whereas the extrapulmonary, associated and disseminated forms were predominantly seen in patients with a more severe level of immunosuppression.
- The TB recovery rate in HIV infected patients was similar to the expected rate in non-infected individuals. The best results were obtained when regimens containing rifampicin were used.
- The pancytopenia and the low levels of TCD₄⁺ and TCD₈⁺ lymphocytes at the initial time point of the study correlate with a unfavorable outcome of the TB infection, and therefore they can be considered potential prognostic factors. However, the values of TCD₈⁺ lymphocyte counts were the most important variable.

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